detecting an analyte of Test No. 10 is 0.68 nA. From these results, it is found that the photocurrent detected by the method for electrochemically detecting an analyte of Test No. 9 is larger than the photocurrent detected by the method for electrochemically detecting an analyte of Test No. 10.

[0323] The above results suggest that the analyte can be detected with higher sensitivity by using a multivalent-labeled binding substance in which more labeling substances are immobilized to the binding substance through the interaction between modulators in order to label the analyte.

Example 2-4

[0324] Quantitative detection of mouse IgG with multivalent-labeled DNA

(1-1) Trapping of Analyte

[0325] Silicone rubber (0.1 mm in thickness) was placed around the working electrode of the working electrode substrate obtained in Preparation example 2-2 so that a partition was formed. Thereafter, TBS-T containing 0.4% by mass of Block Ace [manufactured by DS Pharma Biomedical Co., Ltd.] was poured into the space surrounded by the working electrode substrate and the silicone rubber. Thereafter, the working electrode substrate was incubated at 25° C. for 30 minutes. After the washing of the working electrode substrate with TBS-T, 30 μL of TBS-T containing 1% by mass of BSA containing 10 ng/mL mouse IgG (analyte) was added to the above space. Thereafter, the working electrode substrate was incubated at 25° C. for 1 hour to allow the analyte [mouse IgG] to be trapped by the trapping substance [anti-mouse IgG $F(ab')_2$ antibody].

(1-2) Labeling

[0326] The working electrode substrate subjected to the process (1-1) was washed with TBS-T. Then, biotin-labeled anti-mouse IgG $F(ab')_2$ antibody [manufactured by Dako] was added to TBS-T containing 1% by mass of BSA so that the concentration of the antibody was 4 μg/mL. Thereafter, 30 μL of the obtained mixture was poured into the above space. Thereafter, the working electrode substrate was incubated at 25° C. for 1 hour. Thus, the biotin-labeled anti-mouse IgG $F(ab')_2$ antibody (the first conjugate) was bound to the analyte trapped by the trapping substance. The biotin-labeled anti-mouse IgG $F(ab')_2$ antibody was obtained by labeling the anti-mouse IgG $F(ab')_2$ antibody with biotin.

[0327] Then, the working electrode substrate was washed with TBS-T. Thereafter, 2 mg/mL of streptoavidin[manufactured by Vector Laboratories] (the second conjugate) was added to TBS-T so that its concentration was 4 μg/mL. 30 μL of the obtained mixture was poured into the above space. Thereafter, the working electrode substrate was incubated at 25° C. for 30 minutes. Thus, streptoavidin was bound to the biotin-labeled anti-mouse IgG $F(ab')_2$ antibody on the working electrode.

(1-3) Dye-Labeling

[0328] The working electrode substrate subjected to the process (1-2) was washed with TBS-T. Then, TBS-T was added to 30 μL of a solution containing the biotinylated-DNA/Alexa Fluor 750-labeled DNA complex obtained in Preparation example 2-5 (concentration of the complex: 93 μg/mL) in an amount 10 times the amount of the solution. Thereafter, 30 μL of the obtained mixture was poured into the above space. Thereafter, the working electrode substrate was incubated at 25° C. for 30 minutes. Thus, a complex containing the analyte, the first conjugate, the second conjugate, and the labeled form was formed on the working electrode. The complex formed of the first conjugate and the second conjugate corresponds to the binding substance in the label binding substance obtained in Example 2-1. That is, on the working electrode, Alexa Fluor 750 as the labeling substance is bound to the binding substance bound to the analyte via DNA as the modulator.

(2) Measurement of Photocurrent

[0329] Silicone rubber was placed around the working electrode substrate so that a 0.2-mm-thick side wall was formed. Then, the space surrounded by the working electrode substrate and the silicone rubber was filled with the electrolytic solution obtained in Preparation example 2-3. The space filled with the electrolytic solution was sealed with the counter electrode substrate obtained in Preparation example 2-4 from the upper side of the working electrode substrate. Thus, the working electrode and the counter electrode are brought into contact with the electrolytic solution. Then, the detection chip including the working electrode substrate and the counter electrode was placed in an electrochemical measurement device. The working electrode lead and the counter electrode lead were connected to the ammeter.

[0330] The light source (wavelength: 781 nm, laser light source with an output power of 13 mW) emits excitation light from the working electrode substrate side toward the counter electrode substrate. The labeling substance Alexa Fluor 750 is excited by photoirradiation, thereby generating electrons. When the generated electrons are transported to the working electrode, current flows between the working electrode and the counter electrode. Then, the electric current was measured.

[0331] The current was measured by performing the same operation as described above except that 100 pg/mL of mouse IgG and 1 ng/mL of mouse IgG were used as the analyte in place of 10 ng/mL of mouse IgG in (1-1). The operation was performed in the same manner as described above except that the analyte was not used. The control experiment when the analyte was not present was performed.

[0332] FIG. **36** is a graph showing examined results of a relationship between the concentration of the analyte (mouse IgG) and photocurrent in Example 2-4.

[0333] From the results shown in FIG. **36**, it is found that the photocurrents detected when the concentrations of the analyte is 100 pg/mL, 1 ng/mL, and 10 ng/mL are 0.95 nA, 2.85 nA, and 14.9 nA, respectively. It is found that the photocurrent detected when the analyte is not present is 0.47 nA. From these results, it is found that the photocurrent detected by the method for electrochemically detecting an analyte is increased according to the concentration of the analyte. From these results, it is found that when the concentration of the analyte is in the range of 100 pg/mL to 10 ng/mL, the analyte can be quantitatively detected.

[0334] The above results suggest that the analyte can be quantitatively detected with high sensitivity by using a multivalent-labeled binding substance in which more labeling substances are immobilized to the binding substance through the interaction between modulators in order to label the analyte.

Preparation Example 2-6

Production of Working Electrode Substrate

[0335] A working electrode body composed of a thin film (about 200 nm in thickness) of tin-doped indium oxide was